

### AMENDMENT TO THE CLAIMS

This listing of claims will replace all prior versions, and listing, of claims in the application:

Please amend the claims as follows:

#### Listing of Claims:

Claim 1 (withdrawn - currently amended): An isolated, synthetic or recombinant nucleic acid comprising

(a) a nucleic acid sequence having at least ~~80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%~~, 96%, 97%, 98%, 99%, or more or complete (100%) sequence identity to SEQ ID NO:37, ~~or nucleic acids encoding an enzymatically active fragment thereof~~, wherein the nucleic acid encodes at least one polypeptide having a glucanase activity the polypeptide of claim 60;

~~(b) the nucleic acid sequence of (a), wherein the sequence identities are determined by analysis with a sequence comparison algorithm or by a visual inspection;~~

~~(c) the nucleic acid sequence of (b), wherein the sequence comparison algorithm comprises a BLASTN program using as defaults a wordlength (W) of 11, an expectation (E) of 10, M=5, N=4 and a comparison of both strands, and all other options are set to default;~~

~~(d) a nucleic acid sequence encoding a polypeptide having a sequence as set forth in SEQ ID NO:38, or enzymatically active fragments thereof;~~

[[~~(e)~~]](b) a nucleic acid sequence that hybridizes under stringent conditions to a complement of a nucleic acid comprising SEQ ID NO:37, wherein the nucleic acid encodes [[a]]the polypeptide of claim 60 having a glucanase activity and the stringent conditions include a wash step comprising a wash in 0.2X SSC at a temperature of about 65°C for about 15 minutes, ~~and the nucleic acid has at least 80% sequence identity to SEQ ID NO:37;~~

[[~~(f)~~]](c) the nucleic acid of (a), or (b), ~~(c), (d) or (f)~~, encoding a polypeptide having a ~~glucanase activity~~ but lacking a signal sequence and/or a carbohydrate binding module;

[[~~(g)~~]](d) the nucleic acid of (a), (b), or (c), ~~(d), (e) or (f)~~, encoding a polypeptide ~~having glucanase activity~~ and further comprising a heterologous sequence; or

[[~~(h)~~]](e) a nucleic acid fully complementary to the nucleic acid of (a), (b), (c), or (d), ~~(e) or (f)~~.

Claims 2 to 26 (canceled)

Claim 27 (withdrawn – previously presented): A nucleic acid probe for identifying a nucleic acid encoding a polypeptide with a glucanase activity, wherein the probe comprises at least 10 consecutive bases, or at least about 10 to 50, about 20 to 60, about 30 to 70, about 40 to 80, about 60 to 100, or about 50 to 150 consecutive bases, of a sequence comprising (a) SEQ ID NO:37, or (b) the sequence of claim 1, wherein the probe identifies the nucleic acid by binding or hybridization.

Claims 28 to 30 (canceled)

Claim 31 (withdrawn – previously presented): An amplification primer pair for amplifying a nucleic acid encoding a polypeptide having a glucanase activity, wherein the primer pair

(a) is capable of amplifying a nucleic acid comprising the sequence of claim 1 wherein a member of the amplification primer pair comprises an oligonucleotide comprising at least about 10 to 50 consecutive bases of the sequence, or, about 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30 or more consecutive bases of the sequence; or,

(b) comprises a first member having a sequence as set forth by about the first (the 5') 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30 or more residues of the sequence of claim 1, and a second member having a sequence as set forth by about the first (the 5') 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30 or more residues of the complementary strand of the first member.

Claims 32 to 33 (canceled)

Claim 34 (withdrawn – previously presented): A glucanase-encoding nucleic acid comprising: a nucleic acid generated by amplification of a polynucleotide using the amplification primer pair of claim 31; (b) the nucleic acid of (a), wherein the amplification is by polymerase chain reaction (PCR); (c) the nucleic acid of (b), wherein the nucleic acid is generated by amplification of a gene library; or (d) the nucleic acid of (c), wherein the gene library is an environmental library.

Claims 35 to 37 (canceled)

Claim 38 (Canceled): ~~An isolated, synthetic or recombinant glucanase, encoded by the glucanase encoding nucleic acid of claim 34.~~

Claim 39 (withdrawn – previously presented): A method of amplifying a nucleic acid encoding a polypeptide having a glucanase activity comprising amplification of a template nucleic acid with an amplification primer sequence pair capable of amplifying the nucleic acid sequence of claim 1.

Claim 40 (withdrawn - previously presented): An expression cassette comprising a nucleic acid comprising the sequence of claim 1.

Claim 41 (withdrawn - previously presented): A vector comprising a nucleic acid comprising the sequence of claim 1.

Claim 42 (withdrawn - previously presented): A cloning vehicle comprising (a) a nucleic acid comprising the sequence of claim 1, wherein the cloning vehicle comprises a viral vector, a plasmid, a phage, a phagemid, a cosmid, a fosmid, a bacteriophage or an artificial chromosome; (b) the cloning vehicle of (a), wherein the viral vector comprises an adenovirus vector, a retroviral vector or an adeno-associated viral vector; (c) the cloning vehicle of (b), comprising a bacterial artificial chromosome (BAC), a plasmid, a bacteriophage P1-derived vector (PAC), a yeast artificial chromosome (YAC), or a mammalian artificial chromosome (MAC).

Claims 43 and 44 (canceled)

Claim 45 (withdrawn - previously presented): A transformed cell comprising (a) a nucleic acid comprising the sequence of claim 1; (b) an expression cassette as set forth in claim 40; or (c) the transformed cell of (a) or (b), wherein the cell is a bacterial cell, a mammalian cell, a fungal cell, a yeast cell, an insect cell or a plant cell.

Claims 46 and 47 (canceled)

Claim 48 (withdrawn – previously presented): A transgenic non-human animal comprising (a) the sequence of claim 1; or (b) the transgenic non-human animal of (a), wherein the animal is a mouse.

Claim 49 (canceled)

Claim 50 (withdrawn – previously presented): A transgenic plant comprising (a) the sequence of claim 1; or, (b) the transgenic plant of (a), wherein the plant is a corn plant, a sorghum plant, a potato plant, a tomato plant, a wheat plant, an oilseed plant, a rapeseed plant, a soybean plant, a rice plant, a barley plant, a grass, or a tobacco plant.

Claim 51 (canceled)

Claim 52 (withdrawn – previously presented): A transgenic seed comprising (a) the sequence of claim 1; or, (b) the transgenic seed of (a), wherein the seed is a corn seed, a wheat kernel, an oilseed, a rapeseed, a soybean seed, a palm kernel, a sunflower seed, a sesame seed, a rice, a barley, a peanut or a tobacco plant seed.

Claim 53 (canceled)

Claim 54 (withdrawn - previously presented): An antisense oligonucleotide comprising (a) a nucleic acid sequence complementary to or capable of hybridizing under stringent conditions to the sequence of claim 1; or, (b) the antisense oligonucleotide of (a), wherein the antisense oligonucleotide is between about 10 to 50, about 20 to 60, about 30 to 70, about 40 to 80, or about 60 to 100 bases in length.

Claim 55 (canceled)

Claim 56 (withdrawn – previously presented): A method of inhibiting the translation of a glucanase-message in a cell comprising administering to the cell or expressing in the cell

an antisense oligonucleotide comprising a nucleic acid sequence complementary to or capable of hybridizing under stringent conditions to the sequence of claim 1.

Claim 57 (withdrawn - previously presented): A double-stranded inhibitory RNA (RNAi) molecule comprising (a) a subsequence of the sequence of claim 1; or, (b) the sequence of (a), wherein the RNAi is about 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25 or more duplex nucleotides in length.

Claim 58 (canceled)

Claim 59 (withdrawn - previously presented): A method of inhibiting the expression of a glucanase in a cell comprising administering to the cell or expressing in the cell a double-stranded inhibitory RNA (iRNA), wherein the RNA comprises a subsequence of the sequence of claim 1.

Claim 60 (currently amended): An isolated, synthetic, or recombinant polypeptide comprising:

(i) ~~having~~ an amino acid sequence at least ~~80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or more or~~ complete (100%) sequence identity to SEQ ID NO:38, or an enzymatically active fragment thereof, wherein the polypeptide or the enzymatically active fragment has a glucanase activity;

(ii) ~~the amino acid sequence of (i), wherein the sequence identities are determined by analysis with a sequence comparison algorithm or by a visual inspection;~~

(iii) ~~the nucleic acid sequence of (b), wherein the sequence comparison algorithm comprises a BLAST version 2.2.2 algorithm having a filtering setting set to blastall -p blastp -d "nr\_pataa" -F F, and all other options are set to default;~~

~~[[iv]]~~ (ii) an amino acid sequence encoded by [[the]] a nucleic acid of claim 1 having at least 95% sequence identity to SEQ ID NO.: 37, wherein the polypeptide has glucanase activity;

~~(v) the amino acid sequence of (i), (ii), (iii) or (iv) and having at least one conservative amino acid residue substitution and retaining a glucanase activity, wherein a~~

~~conservative substitution comprises substituting an amino acid residue by another amino acid of like characteristics;~~

~~(vi) the amino acid sequence of (v), wherein the conservative substitution comprises replacement of an aliphatic amino acid with another aliphatic amino acid; or, replacement of a Serine with a Threonine or vice versa; or, replacement of an acidic residue with another acidic residue; or, replacement of a residue bearing an amide group with another residue bearing an amide group; or, exchange of a basic residue with another basic residue; or, replacement of an aromatic residue with another aromatic residue, or a combination thereof;~~

~~(vii) the amino acid sequence of (vi), wherein the aliphatic residue comprises Alanine, Valine, Leucine, Isoleucine or a synthetic equivalent thereof; or, the acidic residue comprises Aspartic acid, Glutamic acid or a synthetic equivalent thereof; or, the residue comprising an amide group comprises Aspartic acid, Glutamic acid or a synthetic equivalent thereof; or, the basic residue comprises Lysine, Arginine or a synthetic equivalent thereof; or, the aromatic residue comprises Phenylalanine, Tyrosine or a synthetic equivalent thereof;~~

~~[[viii]] (iii) the amino acid sequence of (i), or (ii), (iii), (iv), (v), (vi) or (vii), having a glucanase activity but lacking a signal sequence and/or a carbohydrate binding module~~

~~[[ix]] (iv) the amino acid sequence of (i), (ii), or (iii), (iv), (v), (vi), (vii) or (viii), further comprising a heterologous sequence; or~~

~~[[x]] (v) the amino acid sequence of (i), (ii), (iii), (iv), or (v), (vi), (vii), (viii), or (ix), wherein the polypeptide comprises at least one glycosylation site; or, wherein the polypeptide comprises at least one N-linked glycosylation site, or, wherein the polypeptide is glycosylated after being expressed in a *P. pastoris* or a *S. pombe*.~~

Claims 61 to 90 (canceled)

Claim 91 (previously presented): A protein preparation comprising the polypeptide of claim 60, wherein the protein preparation comprises a liquid, a solid or a gel.

Claim 92 (canceled): ~~A heterodimer comprising:~~

~~(a) the polypeptide of claim 60 and a second domain;~~

~~(b) the heterodimer of (a), wherein the second domain is a polypeptide and the heterodimer is a fusion protein; or~~

~~(c) the heterodimer of (a) or (b), wherein the second domain is an epitope or a tag.~~

Claims 93 to 94 (canceled)

Claim 95 (canceled): ~~A homodimer comprising the polypeptide of claim 60.~~

Claim 96 (previously presented): An immobilized polypeptide, wherein the polypeptide comprises: (a) the sequence of claim 60; or, (b) the immobilized polypeptide of (a), wherein the polypeptide is immobilized on a cell, a metal, a resin, a polymer, a ceramic, a glass, a microelectrode, a graphitic particle, a bead, a gel, a plate, an array or a capillary tube.

Claim 97 (canceled)

Claim 98 (Currently amended): An array comprising[[: (a)]] an immobilized polypeptide ~~comprising the amino acid sequence of claim 60; or (b) an immobilized nucleic acid comprising the nucleic acid sequence of claim 1; or (c) a combination thereof.~~

Claim 99 (canceled)

Claim 100 (withdrawn - previously presented): An isolated, synthetic or recombinant antibody (a) that specifically binds to the polypeptide of claim 60; or (b) the antibody of (a), wherein the antibody is a monoclonal or a polyclonal antibody.

Claim 101 (canceled)

Claim 102 (previously presented): A hybridoma comprising an antibody that specifically binds to the polypeptide of claim 60.

Claim 103 (withdrawn – previously presented): A method of isolating or identifying a polypeptide with a glucanase activity comprising:

- (a) providing the antibody of claim 100;
- (b) providing a sample comprising polypeptides; and

(c) contacting the sample of (b) with the antibody of (a) under conditions wherein the antibody can specifically bind to the polypeptide, thereby isolating or identifying a polypeptide having a glucanase activity.

Claim 104 (withdrawn – previously presented): A method of making an anti-glucanase antibody comprising (a) administering to a non-human animal the nucleic acid of claim 1 in an amount sufficient to generate a humoral immune response, thereby making an anti-glucanase antibody; or, (b) administering to a non-human animal the polypeptide of claim 60 in an amount sufficient to generate a humoral immune response, thereby making an anti-glucanase antibody.

Claim 105 (canceled)

Claim 106 (withdrawn – previously presented): A method of producing a recombinant polypeptide comprising:

(A) (a) providing the nucleic acid of claim 1; and (b) expressing the nucleic acid of (a) under conditions that allow expression of the polypeptide, thereby producing a recombinant polypeptide; or

(B) the method of (A), further comprising transforming a host cell with the nucleic acid of (A) (a) followed by expressing the nucleic acid of (A) (a), thereby producing a recombinant polypeptide in a transformed cell.

Claim 107 (canceled)

Claim 108 (withdrawn – previously presented): A method for identifying a polypeptide having a glucanase activity comprising:

(a) providing the polypeptide of claim 60;

(b) providing a glucanase substrate; and

(c) contacting the polypeptide with the substrate of (b) and detecting a decrease in the amount of substrate or an increase in the amount of a reaction product, wherein a decrease in the amount of the substrate or an increase in the amount of the reaction product detects a polypeptide having a glucanase activity.



Claim 109 (withdrawn – previously presented): A method for identifying a glucanase substrate comprising:

- (a) providing the polypeptide of claim 60;
- (b) providing a test substrate; and
- (c) contacting the polypeptide of (a) with the test substrate of (b) and detecting a decrease in the amount of substrate or an increase in the amount of reaction product, wherein a decrease in the amount of the substrate or an increase in the amount of a reaction product identifies the test substrate as a glucanase substrate.

Claim 110 (withdrawn – previously presented): A method of determining whether a test compound specifically binds to a polypeptide comprising:

- (a) expressing a nucleic acid or a vector comprising the nucleic acid under conditions permissive for translation of the nucleic acid to a polypeptide, wherein the nucleic acid has the sequence of claim 1;
- (b) providing a test compound;
- (c) contacting the polypeptide with the test compound; and
- (d) determining whether the test compound of (b) specifically binds to the polypeptide.

Claim 111 (withdrawn – previously presented): A method of determining whether a test compound specifically binds to a polypeptide comprising:

- (a) providing the polypeptide of claim 60;
- (b) providing a test compound;
- (c) contacting the polypeptide with the test compound; and
- (d) determining whether the test compound of (b) specifically binds to the polypeptide.

Claim 112 (withdrawn – previously presented): A method for identifying a modulator of a glucanase activity comprising:

- (A) (a) providing the polypeptide of claim 60;
- (b) providing a test compound;
- (c) contacting the polypeptide of (a) with the test compound of (b) and measuring an activity of the glucanase, wherein a change in the glucanase in the presence of the test

compound compared to the activity in the absence of the test compound provides a determination that the test compound modulates the glucanase activity; or

(B) the method of (A), wherein the glucanase activity is measured by providing a glucanase substrate and detecting a decrease in the amount of the substrate or an increase in the amount of a reaction product, or, an increase in the amount of the substrate or a decrease in the amount of a reaction product; or

(C) the method of (B), wherein a decrease in the amount of the substrate or an increase in the amount of the reaction product with the test compound as compared to the amount of substrate or reaction product without the test compound identifies the test compound as an activator of a glucanase activity; or, wherein an increase in the amount of the substrate or a decrease in the amount of the reaction product with the test compound as compared to the amount of substrate or reaction product without the test compound identifies the test compound as an inhibitor of a glucanase.

Claims 113 to 125 (canceled)

Claim 126 (withdrawn – previously presented): A method for isolating or recovering a nucleic acid encoding a polypeptide with a glucanase activity from an environmental sample comprising:

(A) (a) providing the amplification primer pair of claim 31;

(b) isolating a nucleic acid from the environmental sample or treating the environmental sample such that nucleic acid in the sample is accessible for hybridization to the amplification primer pair; and,

(c) combining the nucleic acid of [[step]] (b) with the amplification primer pair of (a) and amplifying nucleic acid from the environmental sample, thereby isolating or recovering a nucleic acid encoding a polypeptide with a glucanase activity from an environmental sample;

(B) the method of (A), wherein each member of the amplification primer pair comprises an oligonucleotide comprising at least about 10 to 50, or about 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30 or more consecutive bases of, the sequence of claim 1;

(C) (a) providing a polynucleotide probe comprising the sequence of claim 1;

(b) isolating a nucleic acid from the environmental sample or treating the environmental sample such that nucleic acid in the sample is accessible for hybridization to a polynucleotide probe of (a);

(c) combining the isolated nucleic acid or the treated environmental sample of (b) with the polynucleotide probe of (a); and

(d) isolating a nucleic acid that specifically hybridizes with the polynucleotide probe of (a), thereby isolating or recovering a nucleic acid encoding a polypeptide with a glucanase activity from an environmental sample; or

(D) the method of (A), (B) or (C), wherein the environmental sample comprises a water sample, a liquid sample, a soil sample, an air sample or a biological sample, or, wherein the biological sample is derived from a bacterial cell, a protozoan cell, an insect cell, a yeast cell, a plant cell, a fungal cell or a mammalian cell.

Claims 127 to 130 (canceled)

Claim 131 (withdrawn – previously presented): A method of generating a variant of a nucleic acid encoding a polypeptide with a glucanase activity comprising:

(A) (a) providing a template nucleic acid comprising the sequence of claim 1; and  
(b) modifying, deleting or adding one or more nucleotides in the template sequence, or a combination thereof, to generate a variant of the template nucleic acid; or

(B) the method of (A), further comprising expressing the variant nucleic acid to generate a variant glucanase polypeptide;

(C) the method of claim (A) or (B), wherein the modifications, additions or deletions are introduced by a method comprising error-prone PCR, shuffling, oligonucleotide-directed mutagenesis, assembly PCR, sexual PCR mutagenesis, *in vivo* mutagenesis, cassette mutagenesis, recursive ensemble mutagenesis, exponential ensemble mutagenesis, site-specific mutagenesis, gene reassembly, Gene Site Saturation Mutagenesis™ (GSSM™), synthetic ligation reassembly (SLR), recombination, recursive sequence recombination, phosphothioate-modified DNA mutagenesis, uracil-containing template mutagenesis, gapped duplex mutagenesis, point mismatch repair mutagenesis, repair-deficient host strain mutagenesis, chemical mutagenesis, radiogenic mutagenesis, deletion mutagenesis, restriction-selection mutagenesis, restriction-purification mutagenesis, artificial gene

synthesis, ensemble mutagenesis, chimeric nucleic acid multimer creation, or any combination thereof; or

(D) the method of (A) or (B) iteratively repeated until a glucanase an altered or different activity or an altered or different stability from that of a polypeptide encoded by the template nucleic acid is produced;

(E) the method of (A), (B), (C) or (D), wherein the variant glucanase polypeptide is thermotolerant, and retains some activity after being exposed to an elevated temperature;

(F) the method of (A), (B), (C) or (D), wherein the variant glucanase polypeptide has increased glycosylation as compared to the glucanase encoded by a template nucleic acid;

(G) the method of (A), (B), (C) or (D), wherein the variant glucanase polypeptide has a glucanase activity under a high temperature, wherein the glucanase encoded by the template nucleic acid is not active under the high temperature;

(H) the method of (A), (B), (C) or (D), wherein the method is iteratively repeated until a glucanase coding sequence having an altered codon usage from that of the template nucleic acid is produced; or

(I) the method of (A), (B), (C) or (D), wherein the method is iteratively repeated until a glucanase gene having higher or lower level of message expression or stability from that of the template nucleic acid is produced.

Claims 132 to 140 (canceled)

Claim 141 (withdrawn – previously presented): A method for modifying codons in a nucleic acid encoding a polypeptide with a glucanase activity to increase or decrease its expression in a host cell, the method comprising:

(A) (a) providing a nucleic acid encoding a polypeptide with a glucanase activity comprising a sequence as set forth in claim 1; and,

(b) identifying a non-preferred or a less preferred codon in the nucleic acid of (a) and replacing it with a preferred or neutrally used codon encoding the same amino acid as the replaced codon, wherein a preferred codon is a codon over-represented in coding sequences in genes in the host cell and a non- preferred or less preferred codon is a codon under-represented in coding sequences in genes in the host cell, thereby modifying the nucleic acid to increase its expression in a host cell;

(B) (a) providing a nucleic acid encoding a polypeptide with a glucanase activity comprising the sequence of claim 1; and,

(b) identifying a codon in the nucleic acid of (a) and replacing it with a different codon encoding the same amino acid as the replaced codon, thereby modifying codons in a nucleic acid encoding a glucanase; or,

(C) (a) providing a nucleic acid encoding a glucanase polypeptide comprising the sequence of claim 1; and,

(b) identifying a non-preferred or a less preferred codon in the nucleic acid of [[step]] (a) and replacing it with a preferred or neutrally used codon encoding the same amino acid as the replaced codon, wherein a preferred codon is a codon over-represented in coding sequences in genes in the host cell and a non- preferred or less preferred codon is a codon under-represented in coding sequences in genes in the host cell, thereby modifying the nucleic acid to increase its expression in a host cell;

(D) (a) providing a nucleic acid encoding a glucanase polypeptide comprising the sequence of claim 1; and

(b) identifying at least one preferred codon in the nucleic acid of (a) and replacing it with a non-preferred or less preferred codon encoding the same amino acid as the replaced codon, wherein a preferred codon is a codon over-represented in coding sequences in genes in a host cell and a non- preferred or less preferred codon is a codon under-represented in coding sequences in genes in the host cell, thereby modifying the nucleic acid to decrease its expression in a host cell; or,

(E) the method of (A), (B), (C) or (D), wherein the host cell is a bacterial cell, a fungal cell, an insect cell, a yeast cell, a plant cell or a mammalian cell.

Claims 142 to 145 (canceled)

Claim 146 (withdrawn – currently amended): A method for producing a library of nucleic acids encoding a plurality of modified glucanase active sites or substrate binding sites, wherein the modified active sites or substrate binding sites are derived from a first nucleic acid comprising a sequence encoding a first active site or a first substrate binding site the method comprising:

(A) (a) providing a first nucleic acid encoding a first active site or first substrate binding site, wherein the first nucleic acid sequence comprises a sequence that hybridizes

under stringent conditions to the sequence of claim 1, and the nucleic acid encodes a glucanase active site or a glucanase substrate binding site;

(b) providing a set of mutagenic oligonucleotides that encode naturally-occurring amino acid variants at a plurality of targeted codons in the first nucleic acid; and,

(c) using the set of mutagenic oligonucleotides to generate a set of active site-encoding or substrate binding site-encoding variant nucleic acids encoding a range of amino acid variations at each amino acid codon that was mutagenized, thereby producing a library of nucleic acids encoding a plurality of modified glucanase active sites or substrate binding sites;

(B) the method of (A), comprising mutagenizing the first nucleic acid of (a) by a method comprising an optimized directed evolution system, Gene Site-Saturation Mutagenesis<sup>[[TM]]</sup> (GSSM<sup>[[TM]]</sup>), or a synthetic ligation reassembly (SLR); or

(C) the method of (A), comprising mutagenizing the first nucleic acid of (a) or variants by a method comprising error-prone PCR, shuffling, oligonucleotide-directed mutagenesis, assembly PCR, sexual PCR mutagenesis, in vivo mutagenesis, cassette mutagenesis, recursive ensemble mutagenesis, exponential ensemble mutagenesis, site-specific mutagenesis, gene reassembly, Gene Site-Saturation Mutagenesis<sup>[[TM]]</sup> (GSSM<sup>[[TM]]</sup>), synthetic ligation reassembly (SLR), recombination, recursive sequence recombination, phosphothioate-modified DNA mutagenesis, uracil-containing template mutagenesis, gapped duplex mutagenesis, point mismatch repair mutagenesis, repair-deficient host strain mutagenesis, chemical mutagenesis, radiogenic mutagenesis, deletion mutagenesis, restriction-selection mutagenesis, restriction-purification mutagenesis, artificial gene synthesis, ensemble mutagenesis, chimeric nucleic acid multimer creation, or a combination thereof.

Claims 147 to 161 (canceled)

Claim 162 (previously presented): An isolated, synthetic or recombinant signal sequence comprising, or consisting of, the sequence of claim 60 or a sequence as set forth in residues 1 to 14, 1 to 15, 1 to 16, 1 to 17, 1 to 18, 1 to 19, 1 to 20, 1 to 21, 1 to 22, 1 to 23, 1 to 24, 1 to 25, 1 to 26, 1 to 27, 1 to 28, 1 to 28, 1 to 30, 1 to 31, 1 to 32, 1 to 33, 1 to 34, 1 to 35, 1 to 36, 1 to 37, 1 to 38, 1 to 40, 1 to 41, 1 to 42, 1 to 43 or 1 to 44, of SEQ ID NO:38.

Claim 163 (withdrawn – previously presented): A chimeric polypeptide comprising  
(A) at least a first domain comprising signal peptide (SP) having the sequence of claim 162, and at least a second domain comprising a heterologous polypeptide or peptide, wherein the heterologous polypeptide or peptide is not naturally associated with the signal peptide (SP);

(B) the chimeric polypeptide of (A), wherein the heterologous polypeptide or peptide is not a glucanase; or

(C) the chimeric polypeptide of (B), wherein the heterologous polypeptide or peptide is amino terminal to, carboxy terminal to or on both ends of the signal peptide (SP) or a glucanase catalytic domain (CD).

Claim 164 (withdrawn - previously presented): An isolated, synthetic or recombinant nucleic acid encoding a chimeric polypeptide, wherein the chimeric polypeptide comprises at least a first domain comprising signal peptide (SP) having the sequence of claim 162 and at least a second domain comprising a heterologous polypeptide or peptide, wherein the heterologous polypeptide or peptide is not naturally associated with the signal peptide (SP).

Claims 165 to 172 (canceled)

Claim 173 (withdrawn – previously presented): A method for hydrolyzing, breaking up or disrupting a cellulose-, hemicellulose-, lignin-, or glucan-comprising composition comprising:

(A) (a) providing the polypeptide having a glucanase activity of claim 60, or a polypeptide encoded by the nucleic acid of claim 1;

(b) providing a composition comprising a cellulose, hemicellulose, lignin, or glucan; and

(c) contacting the polypeptide of (a) with the composition of (b) under conditions wherein the glucanase hydrolyzes, breaks up or disrupts the cellulose-, hemicellulose-, lignin-, or glucan- comprising composition; or

(B) the method of (A), wherein the composition comprises a biomass, a plant cell, a bacterial cell, a yeast cell, an insect cell, or an animal cell.

Claim 174 (canceled)

Claim 175 (previously presented): A dough or a bread product comprising the polypeptide of claim 60.

Claim 176 (withdrawn - previously presented): A method of dough conditioning comprising contacting a dough or a bread product with at least one polypeptide of claim 60 under conditions sufficient for conditioning the dough.

Claim 177 (previously presented): A beverage comprising the polypeptide of claim 60.

Claim 178 (withdrawn - previously presented): A method of beverage production comprising

(A) administration of at least one polypeptide of claim 60 to a beverage or a beverage precursor under conditions sufficient for decreasing the viscosity of the beverage; or

(B) the method of (A), wherein the beverage or beverage precursor is a wort or a beer.

Claim 179 (canceled)

Claim 180 (previously presented): A food, a feed or a nutritional supplement comprising the polypeptide of claim 60.

Claim 181 (withdrawn - currently amended): A method for utilizing a glucanase nutritional supplement in an animal diet, the method comprising:

(A) preparing a nutritional supplement containing a polypeptide as set forth in claim 60; and administering the nutritional supplement to an animal to increase utilization of a glucan contained in a feed or a food ingested by the animal;

(B) the method of (A), wherein the animal is a human; or

(C) the method of (A), wherein the animal is a ruminant or a monogastric animal.

Claims 182 to 186 (canceled)



Claim 187 (currently amended): An edible enzyme delivery matrix comprising the polypeptide of claim 60 ~~or a polypeptide encoded by the nucleic acid of claim 1.~~

Claim 188 (canceled)

Claim 189 (withdrawn - currently amended): A method for delivering a glucanase to an animal, the method comprising:

(a) preparing an edible enzyme delivery matrix in the form of pellets comprising a granulate edible carrier and a thermostable recombinant glucanase enzyme, wherein the pellets readily disperse the glucanase enzyme contained therein into aqueous media, and administering the edible enzyme delivery matrix to the animal;

(b) the method of (a), wherein the recombinant glucanase enzyme comprises the polypeptide of claim 60 or a polypeptide encoded by the nucleic acid of claim 1;

(c) the method of (a) or (b), wherein the granulate edible carrier comprises a carrier comprising a grain germ, a grain germ that is spent of oil, a hay, an alfalfa, a timothy, a soy hull, a sunflower seed meal, a wheat midd or a combination thereof;

(d) the method of (a), (b) or (c), wherein the edible carrier comprises grain germ that is spent of oil;

(e) the method of (a), (b), (c) or (d), wherein the glucanase enzyme is glycosylated to provide thermostability at pelletizing conditions;

(f) the method of (a), (b), (c), (d) or (e), wherein the delivery matrix is formed by pelletizing a mixture comprising a grain germ and a glucanase;

(g) the method of (a), (b), (c), (d), (e) or (f), wherein the pelletizing conditions include application of steam;

(h) the method of (a), (b), (c), (d), (e), (f) or (g), wherein the pelletizing conditions comprise application of a temperature in excess of about 80°C for about 5 minutes and the enzyme retains a specific activity of at least 350 to about 900 units per milligram of enzyme.

Claims 190 to 196 (canceled)

Claim 197 (withdrawn – previously presented): An isolated, synthetic or recombinant nucleic acid comprising

(a) a sequence encoding a polypeptide having a glucanase activity and a signal sequence, wherein the nucleic acid comprises the sequence of claim 1; or

(b) the nucleic acid of (a), wherein the signal sequence is derived from another glucanase or a non-glucanase enzyme.

Claim 198 (canceled)

Claim 199 (~~canceled~~)~~withdrawn—currently amended~~): ~~An isolated, synthetic or recombinant nucleic acid comprising a sequence encoding a polypeptide having a glucanase activity, wherein the sequence does not contain a signal sequence and the nucleic acid comprises a sequence of claim 1.~~

Claim 200 (previously presented): A cellulose- or cellulose derivative- composition comprising the polypeptide of claim 60.

Claim 201 (previously presented): A wood, wood pulp or wood product comprising the polypeptide of claim 60.

Claim 202 (previously presented): A paper, paper pulp or paper product comprising the polypeptide of claim 60.

Claim 203 (withdrawn - previously presented): A method for reducing lignin in a paper, a wood or wood product comprising contacting the paper, wood or wood product with the polypeptide of claim 60.

Claim 204 (previously presented): A detergent composition comprising a polypeptide as set forth in claim 60.

Claim 205 (previously presented): A pharmaceutical composition comprising the polypeptide of claim 60.

Claim 206 (withdrawn – previously presented): A method for eliminating or protecting animals from a microorganism comprising

- (a) administering the polypeptide of claim 60 to the animal;
- (b) the method of (a), wherein the microorganism is a bacterium; or
- (c) the method of (b), wherein the bacterium is a *Salmonellae*.

Claims 207 to 208 (canceled)

Claim 209 (canceled): ~~A fuel made by the method of claim 210.~~

Claim 210 (withdrawn – previously presented): A method for making a fuel comprising contacting a composition comprising a cellulose, a hemicellulose, a lignin or a glucan with the polypeptide of claim 60.

Claim 211 (previously presented): A dairy product comprising:

- (a) the polypeptide of claim 60; or
- (b) the dairy product of (a), comprising a milk, an ice cream, a cheese or a yogurt.

Claim 212 (canceled)

Claim 213 (withdrawn – previously presented): A method for improving texture and flavor of a dairy product comprising:

- (a) providing the polypeptide of claim 60;
- (b) providing a dairy product; and
- (c) contacting the polypeptide of (a) and the dairy product of (b) under conditions wherein the glucanase can improve the texture or flavor of the dairy product.

Claims 214 to 220 (canceled)

Claim 221 (Currently amended) A composition comprising the polypeptide of claim 60 ~~or the polypeptide encoded by the nucleic acid of claim 1.~~

Claim 222 (Currently amended) A method for oil well drilling, fracturing or treatment comprising use of the polypeptide of claim 60 ~~or the polypeptide encoded by the nucleic acid of claim 1.~~

Claim 223 (new) A polypeptide having glucanase activity comprising an amino acid sequence of SEQ ID NO.: 38.

Claim 224 (new) The polypeptide of claim 222, encoded by the nucleic acid sequence of SEQ ID NO.: 37.